Protecting Farmers' Investment in *Bt* Cotton

PEGGY GREB (K9241-1)

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Close-ups of 12-day-old cotton bollworm larvae on cotton bolls. The smaller larva was fed a diet containing *Bt* proteins, whereas the larger larva was fed a controlled diet. Diet comparisons are used to track the development of *Bt* resistance.

In the past, farmers have used toxic chemicals such as calcium arsenate, Paris green, dichlorodiphenyltrichloroethane (DDT), and toxaphene to rid their cotton crops of insects. Today, however, farmers are buying transgenic seeds that have built-in pest control. ARS researchers are pitching in to ensure that American farmers have made a sound investment and to ensure global competitiveness.

Cotton pests, like pests of many crops, have developed some resistance to many of the insecticides used to control them. Insects persist, almost mockingly, in spite of farmers' fierce attempts to snuff them out.

Tobacco budworm *(Heliothis virescens)* and bollworm *(Helicoverpa zea)* are two of the most destructive pests in cotton and other crops, with costs of control, production, and lost yield of up to $300 million per year in the United States alone. To add to our arsenal of control agents for these and other lepidopteran pests, in the late 1980s industry began to develop crops with built-in pest control from *Bacillus thuringiensis (Bt)* genes, which produce proteins toxic to several insects, including tobacco budworm and bollworm.

Cotton was one of the first crops to benefit from biotechnology-supplied pest protection, and *Bt* cotton is now one of the most widely used transgenic crops. It is currently grown throughout the United States, China, India, and Australia. More than 2 million acres of *Bt* cotton are grown in the United States alone. Other crops, including corn, potatoes, and soybeans, also now contain *Bt* genes.

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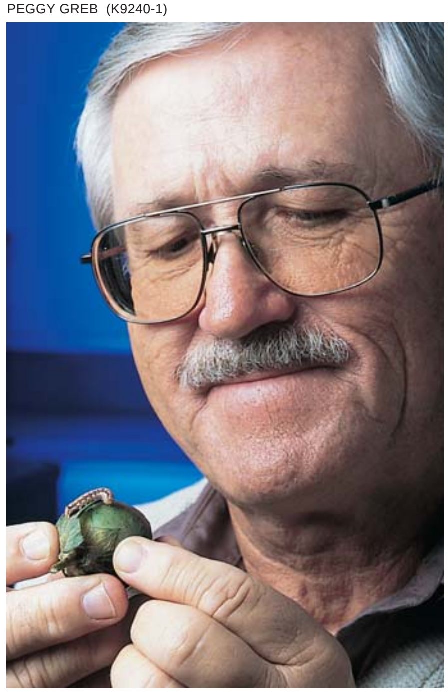

Entomologist D.D. Hardee examines a cotton bollworm larva on a cotton boll.

But *Bt* resistance is cropping up, too. "In addition to synthetic insecticide-resistance development worldwide, several pests have developed resistance to foliar *Bt*, including Indianmeal moths, diamondback moths, and at least nine other insects," says entomologist D.D. Hardee, who heads ARS' Southern Insect Management Research Unit in Stoneville, Mississippi. Because toxins are present in cotton for the entire life of the plant and in all parts of the plant, scientists are concerned that tobacco budworm and bollworm may rapidly develop resistance to *Bt* cotton. Since *Bt* cotton's commercial release in 1996, Hardee's unit has been tracking bollworm and tobacco budworm tolerance to *Bt* proteins as part of a *Bt*-resistance monitoring program.

"Tobacco budworm is the key pest for which *Bt* cotton is used, because it has rapidly developed resistance to previous insecticides. Even though bollworm is more *Bt*-tolerant, it's easily managed with less expensive insecticides and hasn't shown much propensity for developing resistance to insecticides. Nevertheless, we think it's essential that both insects be monitored simultaneously," says Hardee.

"We solicit voluntary participation in our program from all cotton-growing states. Entomologists at universities, other ARS units, and private industry, as well as growers and consultants have responded," says Hardee. "But we need even more people to send us samples because our challenge is always getting enough insects from all areas."

When insect samples arrive in Stoneville, they're sent to ARS entomologist Larry Adams, who raises them through one complete generation on a diet without toxic proteins. The next larval generation from field colonies is then placed on a *Bt* diet and compared with the Stoneville laboratory colony. ARS entomologist Douglas V. Sumerford receives the same colonies for detailed resistance monitoring using a rapid and sensitive assay he developed to help identify pests with tolerance.

"We evaluate the second generation with an artificial diet containing *Bt* protein levels so we can distinguish among larvae in their ability to tolerate *Bt* proteins," says Sumerford. "The more tolerant to *Bt* protein the pest is, the faster it'll grow on the artificial diet and the more likely it'll survive higher doses of *Bt* in the artificial diet. We check the weights of individual larvae, and if a worm is growing fast on the diet, we look for cues indicating a genetic basis for tolerance."

Sumerford is checking for genetic differences between individuals and families. He's developing a genetic map to pinpoint insects that could become resistant to *Bt*. One adult female can lay 400 to 500 eggs, and a moth can fly several miles. Sumerford says, "We want to know if the resistance trait can be spread to other regions. This kind of information combined with the genetic map will help us better monitor any changes in field populations."

"Resistance monitoring is so critical," he adds. "In past years, some cotton growers considered quitting farming, because of the expense involved in controlling tobacco budworm once it developed resistance to pyrethroid pesticides. Our information will help develop remedial action plans for farmers and help preserve the *Bt* technology."

Another part of the monitoring program, carried out by entomologist John J. Adamczyk, Jr., is assessing *Bt* cotton varieties to see how well they control insects. In 1996, only one *Bt* cotton variety was commercially available; today, more than 25 are.

"Many varieties are marketed on their insecticidal qualities, but we want to do further analysis to help growers make a sound investment," says Adamczyk.

He says that by using assays, he and his colleagues have found that some varieties express *Bt* proteins better than others, and certain parts of plants express the proteins better than others. They now want to learn if differences in insect control could also be due to incidents like a farmer's accidentally mixing *Bt* and non-*Bt* seeds or to environmental factors like irrigation or soil salinity. They hope to take high-yielding varieties with the best *Bt* protein levels and incorporate them into a variety-development program.

"We're investigating the next generation of *Bt* cotton, an experimental variety that has two *Bt* genes. It should be commercially available in 2003," says Adamczyk. "Under an experimental use permit from the U.S. Environmental Protection Agency, we are profiling the season-long activity of this double-*Bt*-gene product against various caterpillar pests. Even if there is no cross-resistance between the two *Bt* genes, we still need to make sure that both *Bt* proteins are giving year-round control."

Since the resistance monitoring program started, the Stoneville group hasn't detected any change in the tolerance of tobacco budworm to *Bt* cotton. They must gather data from more areas of the Cotton Belt before they can be certain about bollworm.

"In the meantime, this technology significantly reduces insecticide use," Hardee notes, "and since *Bt* is present in the entire plant for the life of the plant and only affects target pests feeding on the cotton, beneficial insects are left unharmed."

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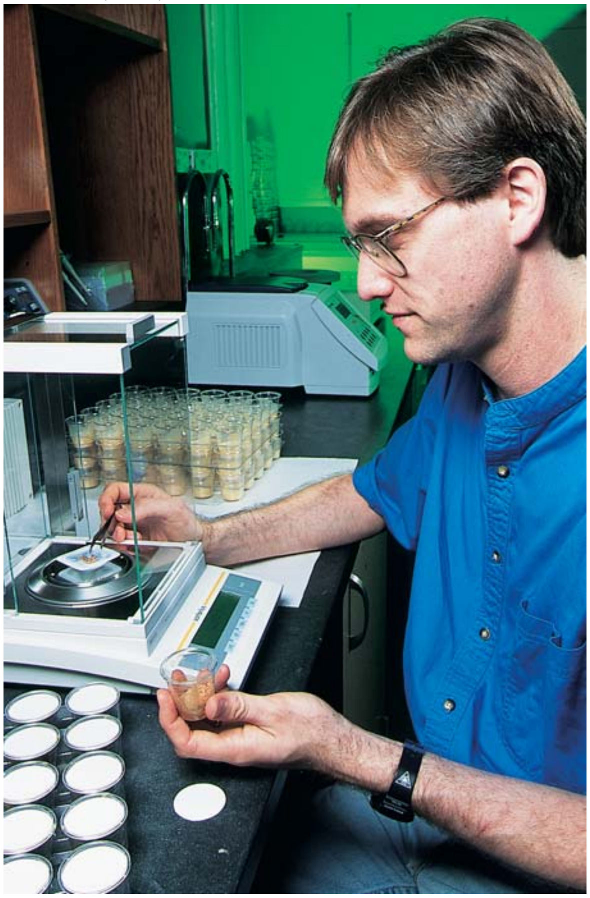

To assess resistance to *Bt* proteins, entomologist Douglas Sumerford weighs a cotton bollworm larva that was exposed to the *Bt*-insecticidal protein Cry1Ac for 12 days.

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Close-up of cotton bollworm larva on the scale.

To attempt to delay the development of *Bt* resistance, federal regulations allow growers to choose one of the following three *Bt*-growing plans:

- Plant a maximum of 80 percent *Bt* cotton along with 20 percent non-*Bt*, with the option of spraying the latter as needed with anything but foliar *Bt* insecticides.
- Plant a maximum of 95 percent *Bt* cotton along with 5 percent non-*Bt*, but the latter cannot be sprayed with any insecticides.
- Plant a maximum of 95 percent *Bt* cotton along with 5 percent non-*Bt*, with the latter planted in the same field with the *Bt* and sprayed only if the 95 percent is sprayed with non-*Bt* foliar insecticides.

"The regulations are an attempt to prolong the effectiveness of *Bt* cotton for decades," says Hardee, "and they are subject to change after the 2001 growing season."

A new publication, "*Bt* Cotton & Management of the Tobacco Budworm-Bollworm Complex," is available from Hardee's group.—By **Tara Weaver-Missick,** ARS.

This research is part of Plant, Microbial, and Insect Genetic Resources, Genomics and Genetic Improvement, an ARS National Program (#301) described on the World Wide Web at http://www.nps.ars.usda.gov.

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